UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

FEB 14 2003

OFFICE OF
PREVENTION, PESTICIDES AND
TOXIC SUBSTANCES

MEMORANDUM:

SUBJECT: Review Closed Bottle Test Data to Assess the Biodegradability of Ucarcide™ P200 Antimicrobial, EPA Reg. No. 464-713, Containing 99.8% 1,2-Benzenedicarboxaldehyde (ortho -phthaladehyde) Preservative

TO: Marshall Swindell, Product Manager, Team 33
Regulatory Management Branch I
Antimicrobials Division (7510C)

FROM: Srinivas Gowda, Microbiologist/Chemist Srinivas Gowda 2/4/03
Risk Assessment and Science Support Branch (RASSB)
Antimicrobials Division (7510C)

THRU: Jonathan Chen, Acting Team Leader, Team One Jonathan Chen 2/11/03
Risk Assessment and Science Support Branch (RASSB)
Antimicrobials Division (7510C)

Norm Cook, Chief Norman J. Cook 02/14/2003
Risk Assessment and Science Support Branch (RASSB)
Antimicrobials Division (7510C)

DP Barcode: D285720
Case: 060726
Submission: S622113
Case Type: 6(a)(2)
Common Name: ortho -pthalaldehyde
Chemical Name:
1,2-Benzenedicarboxaldehyde

EPA Reg. No.: 464-713
MRID No.: 457601-01
Data Submitter: The Dow Chemical Company
PC Code: 129017
CAS#: 643-79-8

INTRODUCTION:

Persuant to Section 6(a)(2) of the Federal Insecticide and Rodenticide Act (FIFRA) as amended, 7 U.S.C. Section 138d(a)(2), the Dow Chemical Company has submitted the Closed Bottle Test Data to establish the Biodegradability of Ucarcide™ P200 Antimicrobial, EPA Reg. No. 464-713, Containing 99.8% 1,2-Benzenedicarboxaldehyde (ortho -phthaladehyde). The submitted Closed Bottle Test Data has undergone review by Srinivas Gowda of Antimicrobials Division's Risk Assessment and Science Support Branch. The registrant used the OCDE/OECD 301D Closed Bottle Test Method to assess the biodegradation of 1,2-Benzenedicarboxaldehyde (ortho -phthaladehyde).

BACKGROUND:

1,2-Benzenedicarboxaldehyde (ortho -phthaladehyde) is an active ingredient in Ucarcide™ P200 Antimicrobial, EPA Reg. No. 464-713. The product is recommended for use by manufacturers in formulating industrial bactericides and preservatives. The submitted study was conducted to determine the percent biodegradation of the active ingredient, ortho -phthaladehyde.

The amended study entitled "ORTHO-PHTHALDEHYDE: CLOSED BOTTLE TEST" by Edward C. Schaefer and Doug Haberlein, Wildlife International Ltd., 8598 Commerce Drive, Easton, Maryland 21601, Study No. 96U1666, Project No. 142E-102, dated August 13, 2002, has been submitted to the Agency (MRID Number 457601-01) persuant to Section 6(a)(2) of the Federal Insecticide and Rodenticide Act (FIFRA) as amended, 7 U.S.C. Section 138d(a)(2). This study was originally submitted under the MRID Number 457221-01.

METHODOLOGY:

The purpose of this study was to measure dissolved oxygen uptake over a 28-day period and express it as a percentage of the theoretical oxygen demand. The study was conducted according to the procedures specified in the Organization for Economic Cooperation and Development (OCDE/OECD) Guideline for Testing of Chemicals, Guideline 301D and Council of European Communities, Guideline C.4-E, *Closed Bottle*.

The study submitted originally under the MRID Number 457221-01 was amended to include two changes to the protocol employed and resubmitted under the MRID Number 457601-01. The two changes were as follows: (1) the test substance sample code or batch number was changed from TGHO0002 to TGHO002, since the sample code or batch number had been incorrectly entered and (2) the value for the theoretical oxygen demand (2.03 mg/mg) was added to the appendix as it had been inadvertently omitted when the protocol was signed. These amendments had no adverse effect on the outcome or validity of the study.

Apparatus

The test chambers consisted of 300 mL biological oxygen demand (BOD) bottles and were identified by project number, Wildlife International Ltd. test substance ID, test concentration, and bottle number. The test was conducted at a temperature of 20±3°C.

Test Substance

The test substance, ortho-Phthalaldehyde, was received from Union Carbide on October 29, 1996, and was assigned Wildlife International Ltd. identification number 3851. The test substance was a yellow powder and had lot number TGHO 002 with an expiration date of 10/97. The substance was stored in a cabinet at ambient temperature and a sample was analyzed according to GLP Standards. The results of the analysis confirmed the identity of the substance and a purity (% Active Ingredient) of >98 mol%. The calculated theoretical oxygen demand of the test substance was 2.03 mg/mg.

A stock solution of the test substance was prepared at a nominal concentration of 1000 mg/L in Nanopure water. In addition, a stock solution of the reference substance, sodium benzoate, was prepared at a nominal concentration of 1000 mg/L in Nanopure water.

Test Medium

The test medium was a modified biochemical oxygen demand test dilution water and was prepared using Nanopure water. The medium was aerated for approximately 20 minutes and then allowed to stand overnight at test temperature. The dissolved oxygen concentration of the dilution water, prior to use, was 8.8 mg O_2/L.

Test Inoculum

The inoculum was prepared from the supernatant fraction of the secondary clarifier which was collected from Prospect Bay Wastewater Treatment Facility in Grasonville, MD on December 6, 1996. The supernatant fraction was filtered through glass wool to produce the inoculum which was aerated until use. A standard plate count was performed on the inoculum and the plates were incubated at 20±3°C for approximately 48 hours.

Experimental Design

The test consisted of an inoculum control group, a reference group, and a treatment group. Each group contained ten replicate test chambers. The inoculum control was used to measure the dissolved oxygen consumption of the inoculum and was not dosed with a carbon source. The reference chambers were dosed with sodium benzoate, a substance known to be biodegradable, at a concentration of 2 mg/L. The treatment group was dosed with the test substance at a concentration of 2 mg/L. Measurements of oxygen consumption were made on two test chambers from each group on days 0, 7, 14, 21, and 28.

Test Chamber Preparation

The BOD bottles were half filled with the aerated mineral medium. Adequate volumes of test substance and reference substance stock solutions necessary to achieve a concentration of 2 mg/L were added to the appropriate bottles. 0.5 mL of the inoculum was then added to the bottles. The mineral medium was added to each bottle so that all bottles were completely full. The bottles were then stoppered and capped.

Sample Analysis

Dissolved oxygen concentrations were measured using a Yellow Springs Instruments Dissolved Oxygen Meter. Measurements were taken on two test chambers from each of the three groups (control, treatment, and reference) on days 0, 7, 14, 21, and 28. The day zero samples were analyzed immediately after all the bottles had been prepared.

Calculations and Results

Average oxygen uptake was calculated for the three groups (control, reference, and treatment) for each sampling interval.

The BOD was calculated using the following equation:

$$BOD = \frac{(\text{mg } O_2\text{/L uptake test substance - mg } O_2\text{/L uptake blank})}{(\text{mg test substance/L in vessel})}$$

The percent degradation was calculated using the following equation:

$$\% \text{ degradation} = \frac{\text{BOD (mg } O_2\text{/mg test substance)}}{\text{THOD}^a \text{ (mg } O_2\text{/mg test substance)}} * 100$$

[a] THOD = theoretical oxygen demand.

The Study Report stated that the temperature range recorded throughout the test duration was 19 to 20°C and that the standard plate count performed on the inoculum resulted in 4.2×10^5 CFU/mL.

Average oxygen uptake for O-Phthalaldehyde ranged from 0.2 to 1.5 mg/L which translated into a BOD range of 0.0 to 0.7 mg/mg. The percent degradation reached a maximum of 27% by the end of the study duration (Day 28).

Dissolved oxygen uptake, BOD and percent degradation was calculated using the data and equations provided in the Study Report. These results are presented in Table 1 in the Attachment. The theoretical oxygen demand (ThOD) for sodium benzoate was not provided. However, using the information and equations provided in the Study Report, RASSB was able to back-calculate out the ThOD used by the study authors in their calculations. This value was calculated to be 1.67 mg/mg.

SUMMARY OF DATA:

Ten replicate BOD (Biological Oxygen Demand) bottles were filled with aerated mineral medium and samples of the reference (sodium benzoate) or test substance (ortho-phthalaldehyde) and 0.5 mL of inoculum. Dissolved oxygen was measured in each bottle using a Yellow Springs Instruments Dissolved Oxygen Meter. Measurements were taken in each bottle from each of the control, reference, and treatment groups on day 0, 7, 14, 21, and 28.

Percent degradation and BOD were calculated for both the reference substance and the test substance. The BOD was calculated from the initial dissolved oxygen content (mg O_2/L), the dissolved oxygen uptake (mg O_2/L), and the concentration of the test substance in the bottle (mg test substance/L). Percent degradation was calculated from the calculated BOD and

theoretical biological oxygen demand (ThOD), given as 2.03 mg/mg.

Average oxygen uptake ranged from 0.2 to 1.5 mg/L which translated into a BOD range of 0.0 to 0.7 mg/mg. The percent degradation reached a maximum of 27% by the end of the study duration (Day 28).

The test substance, Ortho-Phthalaldehyde, reached a mean degradation of 27% over the 28-day test period. According to calculations using the equations, the BOD at day 28 reached 0.7 mg/mg and the percent degradation was 27%. Degradation did not achieve the 60% value regarded as evidence of ready biodegradability and the product appears to be stable.

RASSB's CONCLUSIONS AND RECOMMENDATIONS:

Risk Assessment and Science Support Branch (RASSB) concludes that the submitted Closed Bottle Test reflects the guidelines specified by the OCDE/OECD 301D Closed Bottle Test Method. The percent degradation of th test substance, ortho-Phthalaldehyde at 2 mg/L was approximately 27%. Degradation did not achieve the 60% value regarded as evidence of ready biodegradability, and the active ingredient, ortho-Phthalaldehyde appears to be stable and could persist in the environment.

cc: Srinivas Gowda/RASSB/AD

Chemical File (129017)/AD

ATTACHMENT:

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Table 1. Average Oxygen Uptake, Biochemical Oxygen Demand (BOD), and Percent Degradation Results

Test Substance	Replicate	Sampling Day	Dissolved Oxygen (mg/L)	Dissolved Oxygen uptake (mg/L)	Average Uptake (mg/L)	BOD (mg/mg)	Percent Degradation (%)
Control	1	0	8.8	0.0	-	-	-
			8.8	0.0			
	2	7	8.4	0.4	0.4	-	-
			8.4	0.4			
	3	14	8.5	0.3	0.3	-	-
			8.5	0.3			
	4	21	8.4	0.4	0.4	-	-
			8.4	0.4			
	5	28	8.4	0.4	0.4	-	-
			8.4	0.4			
Benzoate	1	0	8.8	-	-	-	-
			8.8	-			
	2	7	6.0	2.8	2.9	1.2	73
			5.9	2.9			
	3	14	5.9	2.9	2.9	1.3	78
			5.9	2.9			
	4	21	5.5	3.3	3.3	1.4	85
			5.6	3.2			
	5	28	5.4	3.4	3.5	1.5	91
			5.3	3.5			
O-Phthalaldehyde	1	0	8.8	-	-	-	-
			8.8	-			
	2	7	8.4	0.4	0.4	0.0	0
			8.4	0.4			
	3	14	8.6	0.2	0.2	0.0	0
			8.6	0.2			
	4	21	7.8	1.0	1.0	0.3	15
			7.8	1.0			
	5	28	7.2	1.6	1.5	0.6	27
			7.4	1.4			

Reviewed by: Srinivas Gowda, Microbiologist/Chemist, Team 1 Srinivas Gowda Date 2/4/03

DATA EVALUATION RECORD

STUDY TYPE:	Closed Bottle Test
DP BARCODE:	D285720
PC CODE:	129017
SUBMISSION CODE:	S622750
CASE TYPE:	6(a)(2)
TEST MATERIAL:	1,2-Benzenedicarboxaldehyde
SYNONYMS:	ortho -pthalaldehyde
CITATION:	"ORTHO-PHTHALDEHYDE CLOSED BOTTLE TEST" by Edward C. Schaefer and Doug Haberlein, Wildlife International Ltd., 8598 Commerce Drive, Easton, Maryland 21601, Study No. 96U1666, Project No. 142-102, dated August 13, 2002 (MRID number 457601-01)
SPONSOR:	The Dow Chemical Company